

**DRINKING WATER METHODS APPROVED BY THE U.S. EPA FOR USE UNDER THE TOTAL COLIFORM RULE (TCR)
REPORTED AS PRESENT OR ABSENT – MUST REPORT TOTAL COLIFORM AND EITHER FECAL COLIFORM OR E. COLI**

METHODS	TOTAL COLIFORM Media and Description: Reference Citation:	FECAL COLIFORM Media and Description: Reference Citation:	E. COLI Media and Description: Reference Citation:
FERMENTATION BROTH	LTB (or LB*) , transfer to BGLB broth 1 x 100 ml, 10 x 10 ml, 5 x 20 ml Ref: SM 9221A,B <i>Completed Test not required</i>	EC broth Transfer from LTB Ref: SM 9221E	EC + MUG broth Transfer from LTB Ref: CFR 141.21(f)(6)(i)
	P-A broth (Clark's) , transfer to BGLB broth 1 x 100 ml Ref: SM 9221D	EC broth Transfer from P-A broth Ref: SM 9221E	EC + MUG broth Transfer from P-A broth Ref: CFR 141.21(f)(6)(i)
MEMBRANE FILTRATION	m-Endo or m-Endo LES broth or agar Colonies verified in LTB & BGLB. Total volume tested = 100 ml Ref: SM 9222 A,B,C	EC broth Transfer from m-Endo or m-Endo LES Ref: SM 9221E	EC + MUG broth Transfer from m-Endo or m-Endo LES Ref: CFR 141.21(f)(6)(i)
	MI agar Total volume tested = 100 ml Ref: EPA/600/J-99/225	Not Applicable	Nutrient Agar + MUG agar Transfer membrane from m-Endo or m-Endo LES Ref: CFR 141.21(f)(6)(ii)
	m-ColiBlue24® Total volume tested = 100 ml Ref: from Hach Co.	Not Applicable	m-ColiBlue24® Same plate as for total coliform Ref: from Hach Co.
ENZYME SUBSTRATE	Colilert®, Colilert®18 1 x 100 ml, 10 x 10 ml, Quanti-Tray™, Quanti-Tray™ 2000 Ref: SM 9223, Idexx	Not Applicable	Colilert®, Colilert®18 Same container(s) as for total coliform Ref: SM 9223, Idexx
	Colisure® 1 x 100 ml Ref: from Idexx	Not Applicable	Colisure® Same container(s) as for total coliform Ref: from Idexx
	E*Colite® 1 x 100 ml Ref: from Charm Sciences, Inc.	Not Applicable	E*Colite® Same container as for total coliform Ref: from Charm Sciences, Inc.

* LB (lactose broth) may be used in lieu of LTB (lauryl tryptose broth) if the system conducts 25 parallel tests between these two media and demonstrates that the false-positive and false-negative rate for total coliform, using lactose broth is less than 10%.

SOURCE/SURFACE WATER METHODS APPROVED BY USEPA FOR USE UNDER SURFACE WATER TREATMENT RULE (SWTR) REPORTED AS NUMBER PER 100 ML		
METHOD	TOTAL COLIFORM Media and Description: Reference Citation:	FECAL COLIFORM Media and Description: Reference Citation:
FERMENTATION BROTH	LTB (or LB*), transfer to BGLB broth 5 tubes per 3 dilutions Ref: SM 9221 A,B,C <i>Completed Test Not Required</i>	EC broth Transfer from positive LTB broths Ref: SM 9221E
	Not Applicable	A-1 broth (Direct fecal coliform method) 5 tubes per 3 dilution Ref: SM 9221E
MEMBRANE FILTRATION	m-Endo or m-Endo LES broth or agar Filter volumes to result in membrane with 20-80 coliform colonies & not more than 200 colonies of all types. Colonies verified in LTB & BGLB or by EPA approved rapid test.. Report verified count. Ref. SM 9222 A,B,C	Not Applicable
	Not Applicable	m-FC broth or agar Filter volumes to result in membrane with 20 to 60 fecal colonies. Ref: SM 9222D
	MI agar Ref: EPA/600/J-99/225	Not applicable
ENZYME SUBSTRATE	Colilert[®], Colilert[®] 18 5 tubes per 3 dilutions, Quanti-Tray, Quanti-Tray 2000 Ref: SM 9223, Idexx	Not applicable
METHOD	HETEROTROPHIC BACTERIA	
POUR PLATE	Heterotrophic Plate Count (HPC) Ref: SM 9215B	

* LB (lactose broth may be used in lieu of LTB (lauryl tryptose broth) if the system conducts at least 25 parallel tests between these two media, and this comparison demonstrates that the false positive rate and false negative rate for total coliform using LB is less than 10%.

WASTEWATER METHODS APPROVED BY USEPA FOR USE UNDER CLEAN WATER ACT (CWA) AND REPORTED AS NUMBER PER 100 ML		
METHOD	TOTAL COLIFORM Media and Description: Reference Citation:	FECAL COLIFORM Media and Description: Reference Citation:
FERMENTATION BROTH	LTB , transfer to BGLB broth 5 tubes per 3 dilutions Ref: SM 9221 A,B,C <i>Completed Test required</i>	EC broth Transfer from each positive LTB Ref: SM 9221E
	Not Applicable	A-1 broth (Direct fecal coliform test) 5 tubes per 3 dilutions Ref: SM 9221E
MEMBRANE FILTRATION	m-Endo or m-Endo broth or agar Filter volumes to result in membrane with 20-80 coliform colonies and not more than 200 colonies of all types. Colonies verified in LTB & BGLB or by EPA approved rapid test. Report verified count. Ref: SM 9222 A,B,C	Not Applicable
	Not Applicable	m-FC broth or agar Filter volumes to result in membrane with 20-60 colonies. Ref: SM 9222D
METHOD	FECAL STREPTOCOCCI Media & Description: Reference Citation:	ENTEROCOCCI Media & Description: Reference Citation:
MULTIPLE TUBE TEST	Azide Dextrose broth + Bile Esculin Azide agar ** 5 tubes per 3 dilutions Ref: SM 9230B	Verified with BHI with 6.5% NaCl and BHI at 45°C Transfer positive colonies from BEA agar Ref: 9230B
MEMBRANE FILTRATION	mEnterococcus agar Filter volumes to result in membrane with 20-60 colonies Verification recommended Ref: SM 9230C	Requires additional verification including BHI with 6.5% NaCl and BHI at 45°C. Ref: SM 9230C
	Not applicable	mE agar + EIA substrate Filter volumes to result in membrane with 20-60 colonies Verification recommended Ref: SM 9230C
METHOD	HETEROTROPHIC BACTERIA	
POUR PLATE	Heterotrophic Plate Count (HPC) Ref: SM 9215B	

**Bile Esculin Azide agar is equivalent to PSE agar, with addition of 1 g Na Citrate and 0.1 g Na Azide per liter medium.

RECOMMENDED METHODS FOR THE ANALYSIS OF RECREATIONAL MARINE WATER UNDER AB411 AND REPORTED AS NUMBER PER 100 ML

Lab should read information contained in “Recommended Methods for the Analysis of Recreational Marine Water to Comply with AB 411” on DHS website (www.dhs.ca.gov/ps/ddwem/beaches/AB411_methods.htm) for more specific requirements.

METHOD	TOTAL COLIFORM	FECAL COLIFORM and E. COLI***
	Media and Description: Reference Citation:	Media and Description: Reference Citation:
FERMENTATION BROTH	LTB , transfer to BGLB broth 5 tubes per 3 dilutions Ref: SM 9221 A,B,C <i>Completed Test required.</i>	EC broth Transfer from each positive LTB Ref: SM 9221E
MEMBRANE FILTRATION	m-Endo or m-Endo LES broth or agar Filter volumes to result in membrane with 20-80 coliform colonies and not more than 200 colonies of all types. Colonies verified in LTB & BGLB or by EPA approved rapid test. Report verified count. Ref: SM 9222 A,B,C	Not Applicable
	Not Applicable	m-FC broth or agar Filter volumes to result in membrane with 20-60 colonies. Ref: SM 9222D
ENZYME SUBSTRATE	Colilert 18 100 ml marine water diluted 1:10 with sterile water prior to testing in Quanti-Tray or multiple tube dilution format. Ref: SM 9223	Colilert 18*** Same container as for total coliform. Ref: SM 9223
METHOD	FECAL STREPTOCOCCI	ENTEROCOCCI
	Media & Description: Reference Citation:	Media & Description: Reference Citation:
MEMBRANE FILTRATION	Not applicable	mE agar + EIA substrate Filter volumes to result in membrane with 20-60 colonies. Verification recommended Ref: SM 9230C
	Not applicable	mEI agar Filter volumes to result in membrane with 20-60 colonies. Verification recommended Ref: EPA Method 1600
ENZYME SUBSTRATE	Not applicable	Enterolert 100 ml of marine water initially diluted 1:10 with sterile water prior to testing in Quanti-Tray. Ref: from Idexx

***If E. coli is used as a surrogate for fecal coliforms (Colilert 18) read information contained in “Recommended Methods for the Analysis of Recreational Marine Water to Comply with AB 411” on DHS website (www.dhs.ca.gov/ps/ddwem/beaches/ab411_methods.htm) for more specific requirements.

METHOD REFERENCES

CFR – Code of Federal Regulations, Vol. 40

SM – Standard Methods for the Examination of Water and Wastewater, 18th and 19th ed. for drinking water and 18th ed. for wastewater.

Hach Company, 100 Dayton Avenue, Ames, IA 50010 for description of **m-ColiBlue24[®]**.

IDEXX Laboratories, Inc., One IDEXX Drive, Westbrook, Maine 04092 for description of **Colisure**.

Charm Sciences, Inc., 36 Franklin Street, Malden, MA 02148-4120 for description of **E*Colite[®]**.

Office of Water Resource Center (RC-4100), 401 M. Street SW, Washington D.C. 20460, **EPA/600/J-99/224**, for information on preparation and use of **MI agar**.

GLOSSARY OF SELECTED ACRONYMS:

BGLB	Brilliant Green Lactose Bile broth
BHI	Brain Heart Infusion
CFR	Code of Federal Regulations
CWA	Clean Water Act
EIA	Esculin Iron Agar
EPA	Environmental Protection Agency
LB	Lactose Broth
LTB	Lauryl Tryptose Broth
MUG	4-Methylumbelliferyl-beta-d-glucuronide
PSE	Pfizer Selective Enterococcus Agar
SM	Standard Methods (for the Analysis of Water and Wastewater)
SWTR	Surface Water Treatment Rule
TCR	Total Coliform Rule